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Phylogenetic Relationships and Adaptive Shifts among Major Clades of *Tetragnatha* Spiders (Araneae: Tetragnathidae) in Hawai'i[1]

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ABSTRACT: The role of adaptive shifts in species formation has been the subject of considerable controversy for many years. Here we examine the phylogeny of a large radiation of Hawaiian spiders in the genus *Tetragnatha* to determine the extent to which species splitting is associated with shifts in ecological affinity. We use molecular data from ribosomal 12S and cytochrome oxidase mitochondrial DNA, and allozymes to assess phylogenetic affinity. Ecological associations were recorded for all species under study, and shifts are considered in the context of the phylogeny. Results indicate that there are two major clades of Hawaiian *Tetragnatha,* one of which has abandoned web building (spiny-leg clade), while the other retains the ancestral condition of web building. Within the spiny-leg clade, the molecular information suggests that the species on any one island are generally most closely related to each other. Preliminary results for the web-building "complex" of species indicate that there may be groups of web builders that have speciated in a similar manner. Results of the study suggest that, at least within the spiny-leg clade, matching sets of taxa have evolved independently on the different Hawaiian islands. There appears to have been a one-to-one convergence of the same set of "ecomorph" types on each island in a manner similar to that of lizards of the Caribbean.

THE FORMATION OF new species is a key element in evolutionary advancement and diversification (Mayr 1963). Our research has focused on the pattern and process of speciation in a lineage of spiders in the Hawaiian Islands. The archipelago offers a unique opportunity for examining microevolutionary events culminating in the formation of species, largely because its extreme isolation has allowed repeated and explosive taxonomic diversification from one or a few ancestors (C. Simon 1987). Such rampant speciation is well illustrated in Hawaiian birds (Freed et al. 1987, Tarr and Fleischer 1995), land snails (Cooke et al. 1960), crickets (Otte 1994, Shaw 1995, 1996), and pomace flies (DeSalle 1995, Kaneshiro et al. 1995). However, what makes the Hawaiian archipelago ideal for examination of the process of species formation among such radiations is that it consists of a series of volcanoes arranged within an identifiable chronological framework, ranging from Kaua'i, the oldest of the currently high islands, to Hawai'i, the youngest, largest, and highest island (Carson and Clague 1995). It is therefore reasonable to consider the archipelago as a series of historical snapshots, with population divergence being currently instigated on the youngest island and developed on the successively older islands.

One of the keys to understanding the process of speciation is the elucidation of behavioral or ecological changes associated with species splitting. Such changes frequently may be linked to sexual selection (e.g., Kaneshiro and Giddings 1987). However, where sexual behaviors are less apparent, behavioral or ecological changes at speciation have been most frequently associated with foraging mode (e.g., Grant 1986). For a predator, there are three primary mechanisms

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through which it could shift foraging mode: (1) microhabitat selection, which therefore determines the number and type of prey encountered; (2) behavioral modification in foraging strategy, which becomes tailored to a specific prey type; and (3) morphological modification in feeding structures, which become tailored for capture of a specific prey type. Even the most generalist predators usually exhibit some form of microhabitat selection; the most specialized exhibit morphological and behavioral adaptations to a specific prey type in addition to selecting a microhabitat in which this prey is likely to occur. To what extent are these modifications associated with species formation?

Our study examines changes in ecological affinity associated with species splitting for representatives of a lineage of spiders in the long-jawed orb-weaving genus *Tetragnatha. Tetragnatha* is a large genus, with 295 described species (Platnick 1993). Until recently it was considered "primitive" (Bristowe 1958) within the Araneoidea because of the simplicity of the female genitalia (absence of an epigynum). However, based on characters such as the eyes (showing the derived absence of a tapetum) and other features, it is now recognized that the epigynum has been lost secondarily (see Hormiga et al. [1995] for a list of Tetragnathinae synapomorphies and placement within the family). Use of the chelicerae in mating seems to have precluded the need for an epigynal coupling (Levi 1981). The genus *Tetragnatha* is tremendously abundant and of worldwide distribution (Levi 1981). Traditionally, it has been recognized as one of the most homogeneous genera of spiders, in both morphology (elongate form [Kaston 1948]) and ecology (associated with riparian habitats [Gillespie 1987]).

Based on the collection of R. C. L. Perkins, E. Simon (1900) recognized the speciose nature of the genus *Tetragnatha* in Hawai'i. However, Perkins' spider collection, by his own admission, was incomplete and unrepresentative (Perkins 1913): spiders were collected only in passing during his daylight searching for birds and insects or while he collected insects attracted to a light at night. The majority of endemic Hawaiian spiders are strictly nocturnal and extremely difficult to find during the day (pers. obs.), and they cannot be attracted by lights. Accordingly, the extent of the radiation of *Tetragnatha* in Hawai'i has been uncovered only recently (Gillespie 1991*a,* 1992*a,* 1994), previously being known only from descriptions of a single species by Karsch (1880) and of eight species by E. Simon (1900, redescribed by Okuma [1988, 1990]), the latter based on Perkins' collections. Over the last few years an additional 19 species of Hawaiian *Tetragnatha* have been described (Gillespie 1991*a,* 1992*a,* 1994), and more than 50 new "morphospecies" have been collected (Gillespie and Croom 1995; R.G.G., unpubl. data). This species radiation spans a huge spectrum of colors, shapes, sizes, ecological affinities, and behaviors. Many are web building (plesiomorphic for *Tetragnatha*), with structural modifications of the abdomen that allow concealment within specific microhabitats. Some species have modifications of the cheliceral armature, apparently to allow specialization on specific prey types. One clade of 16 species ("spiny-leg" clade) has abandoned web building, with the concomitant development of long macrosetae along the legs and adoption of a vagile, cursorial predatory strategy (Gillespie 1991*a*).

Phylogenetic analysis of the group to date has revealed that there are three clades of Hawaiian *Tetragnatha* and at least two independent origins (Figure 1 [Gillespie et al. 1994]): (1) The *T. hawaiensis* clade appears to have split earliest from the others. It is a nonspeciose lineage similar in gross morphology to its continental congeners. (2) The spiny-leg clade contains all individuals with long macrosetae on their legs. (3) The remaining taxa comprise a complex of web-building species. The spiny-leg clade and the complex of web-building species may have arisen from one or two separate colonization events. The *T. hawaiensis* clade seems to have arisen from another colonization event. An additional natural colonization seems to have given rise to *Doryonychus raptor,* the sole representative of a genus very similar to *Tetragnatha* (Okuma 1990, Gillespie 1991*b,* 1992*b*). To date, phylogenetic analysis of the spiny-leg clade based on morphological characters has indicated that the most closely related species are on different islands (Gillespie 1993, Gillespie and Croom 1995). "Green" species share a suite of apparent synapomorphies that group them together (Figure 2).

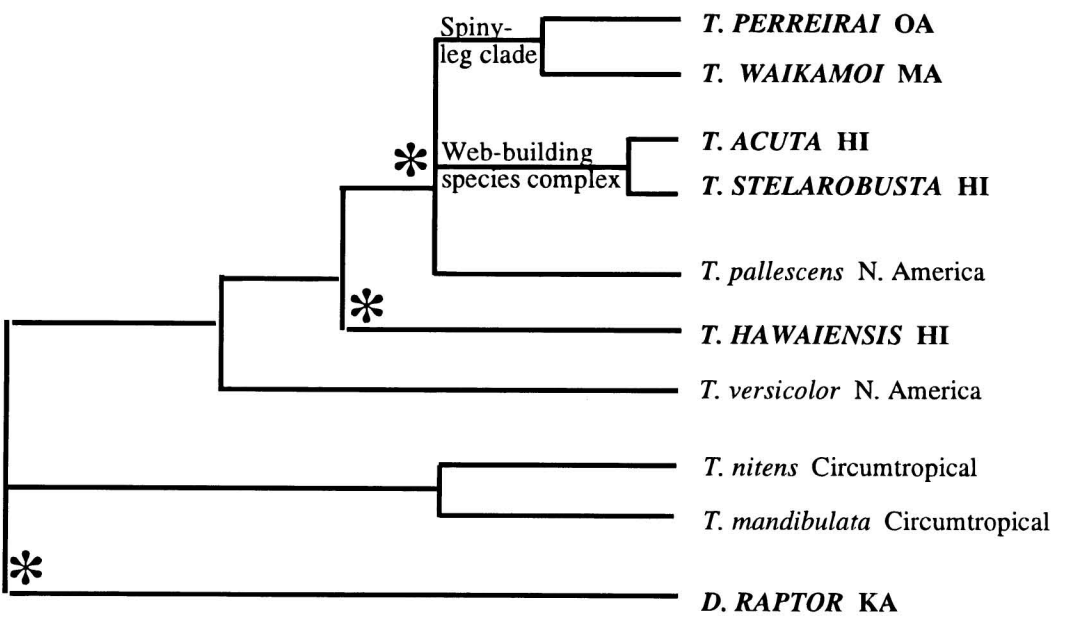

FIGURE 1. Phylogeny of endemic Hawaiian Tetragnathidae relative to representative continental species, showing at least three (two *Tetragnatha,* one *Doryonychus*) introductions into Hawai'i (Gillespie et al. 1994). Maximum likelihood tree based on 12S mtDNA sequences, log likelihood -982.1. Hawaiian taxa shown in bold-faced capital letters. * indicates colonization events. The *T. hawaiensis* clade is 9–13% different from all other clades. Preliminary calculation of sequence differences across all sites shows that each endemic spider is ca. 23% different from *T. mandibulata.* Endemics differ from each other by 3–13% (Gillespie et al. 1994). (Locality abbreviations: Kaua'i, KA; O'ahu, OA; Maui Nui, MA; Hawai'i Island, HI.)

In this study we use molecular characters to make a preliminary phylogenetic assessment of relationships among species in the "spiny-leg" clade, as well as the broad cladistic structure of species groups among the web-building species. We use information from allozymes, and two sets of mitochondrial DNA sequences, 12S and cytochrome oxidase (COI). In addition we examine habitat associations and foraging behavior for the same taxa. By mapping these on to the phylogeny we can assess the role of behavioral or ecological changes in species formation.

MATERIALS AND METHODS

Phylogenetic Analysis

TERMINOLOGY. Many of the Hawaiian *Tetragnatha* are undescribed. Common names only are ascribed to distinct morphotypes. In addition, taxa that are similar in gross morphological appearance are grouped together: (1) "wave" species (such as *T. eurychasma* [Gillespie 1992*a*]) are morphologically similar both to each other and share many features of continental species; (2) "elongate" species (such as *T. stelarobusta* [Gillespie 1992*a*]) are also morphologically very similar to each other.

DNA SEQUENCES. Molecular data were obtained from (1) ribosomal 12S mtDNA, which was amplified and sequenced (using one universal primer and one primer designed specifically for tetragnathid spiders [Croom et al. 1991]) for 10 species in the spiny-leg clade, Kaua'i and Hawai'i representatives of the *T. hawaiensis* clade, and six of the endemic web-building species: *Tetragnatha* sp. "Elongate 5" (like *T. stelarobusta*) (Hawai'i), *T. paludicola* (Maui), *T. trituberculata* (Maui), *T. eurychasma* ("Wave 3") (Maui), *T.* sp. "Wave 2" (like *T. eurychasma*) (O'ahu), *T.* sp. "Golden Dome" (Hawai'i), and *T. acuta* (Hawai'i). We also obtained sequence from the endemic Hawaiian tetragnathid *Doryonychus raptor.* Sequences of spiders over a 204-

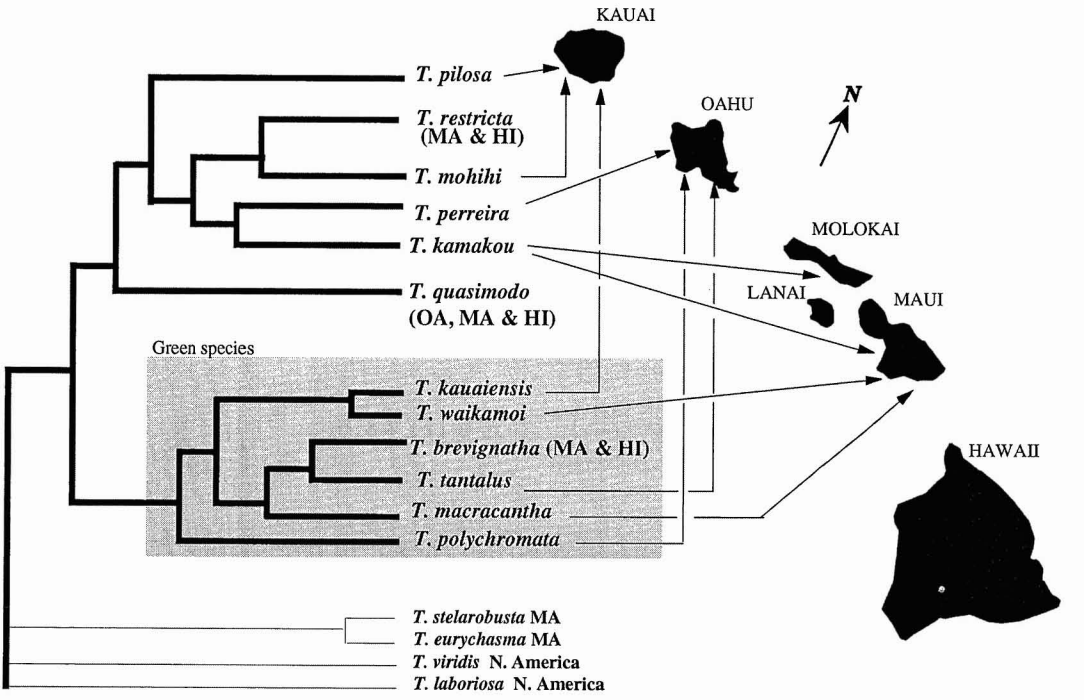

FIGURE 2. Phylogeny of the Hawaiian spiny-leg *Tetragnatha* based on morphological characters (Gillespie 1993). Phylogenetic analysis of 30 morphological characters without weighting gave seven most parsimonious trees (CI 0.52). Subsequent weighting by successive approximations gave the single tree shown of unweighted length 76 (CI 0.73). Arrows point to the location of a species in the archipelago. Shaded area indicates "green" species. (See Figure 1 for locality abbreviations.)

base-pair homologous region were compared, and variation was found at 44 different sites (38 informative). (See Appendix 1 for aligned sequences). (2) Cytochrome oxidase subunit I (COI) was sequenced for six species in the spiny-leg clade (*T. pilosa, T. kauaiensis, T. perreirai, T. quasimodo, T. tantalus,* and *T. brevignatha*), Kaua'i and Hawai'i representatives of the *T. hawaiensis* clade, and 10 of the endemic web-building species: *T.* spp. "Elongate 1," "Elongate 2," and "Elongate 3" (all like *T. stelarobusta*) (O'ahu), *T. paludicola* (Maui), *T.* spp. "Wave 1" and "Wave 2" (both like *T. eurychasma*) (Kaua'i and O'ahu, respectively), *T. filiciphilia* (Maui), *T. acuta* (Maui and Hawai'i populations), *T. albida* (Maui), *T. maka* (Kaua'i), *T.* sp. "Bicolored Jaws" (O'ahu), and *T.* sp. "Long-clawed" (two populations on Hawai'i). The 450-base-pair piece of COI was amplified using primers C1-J-1718 and C1-N-2191 (designed by R. Harrison laboratory [C. Simon et al. 1994]). This region was one codon insertion longer than that of *Drosophila yakuba* (Clary and Wolstenholme 1985). For the range of genetic distances encompassing the major radiation of Hawaiian *Tetragnatha,* both transitions and transversions increased linearly when plotted against Tamura distance (Tamura 1992), suggesting that both transitions and transversions are phylogenetically informative at this level. For the greater distances between the major Hawaiian radiation and species in the "*T. hawaiensis*" clade (separate introduction), transversions are still informative, although transitions show evidence of saturation. Sequences of spiders over a 324-base-pair homologous region were compared and variation was found at 204 different sites (111 informative). (See Appendix 2 for aligned sequences.)

Phylogenies were reconstructed using maximum parsimony (PAUP [Swofford 1993]) with the branch-and-bound search option. Branches

having maximum length zero were collapsed to yield polytomies.

ALLOZYMES. Allozyme frequencies were determined for representatives of the spiny-leg clade using cellulose acetate electrophoresis (Richardson et al. 1986). *Tetragnatha* were homogenized in water and the homogenate applied to cellulose acetate gels (Helena Laboratories, Beaumont, Texas). Electrophoresis was performed at 180v and the gels developed using specific enzyme detection techniques (stain recipes adapted from Hebert and Beaton [1989], buffer recipes from Richardson et al. [1986]). We used seven polymorphic enzyme systems: glyceraldehyde-3-phosphate dehydrogenase (G3PDH, E.C. 1.2.1.12), 6-phosphogluconate dehydrogenase (6PGDH, E.C. 1.1.1.44), and glycerol-3-phosphate dehydrogenase (GPDH, E.C. 1.1.1.8) (buffer for all 0.1 M Tris-citrate, pH 8.2, 1 hr, 1.5 hr, and 0.75 hr, respectively); isocitrate dehydrogenase (IDH, E.C. 1.1.1.42) (buffer of 0.01 M Citrate-phosphate, pH 6.4, 1 hr); phosphoglucose isomerase (PGI, E.C. 5.3.1.9) (buffer of 0.015 M Tris-maleate, pH 7.2, 1 hr); malate dehydrogenase (MDH, E.C. 1.1.1.37) (0.05 M Tris-maleate, pH 7.2, 1 hr); and phosphoglucomutase (PGM, E.C. 2.7.5.1) (0.1 M Tris-maleate, pH 7.8, 1 hr). Between 5 and 30 individuals were sampled from one to five populations of the following species in the "spiny-leg" clade: *T. pilosa* (Kaua'i), *T. kauaiensis* (Kaua'i), *T. quasimodo* (populations from O'ahu, Maui, and Hawai'i), *T. tantalus* (O'ahu, Ko'olau Mountains), *T. polychromata* (O'ahu, Wai'anae Mountains), *T. brevignatha* (populations from Maui and Hawai'i), *T. waikamoi* (Maui), *T. kamakou* (Maui), and *T. restricta* (Maui).

Characteristics of Representative Species

MICROHABITAT ASSOCIATIONS AND FORAGING BEHAVIOR. Each time a spider was collected, its habitat affiliations were noted. For all spiders observed, we noted their activity (in an orb web, hanging in the vegetation, actively running around, or inactive) before capture. Where possible, prey identity was noted subsequent to capture. Detailed behavioral observations were recorded for three sympatric species at 1340 m in Waikamoi, East Maui: *T. stelarobusta, T. quasimodo,* and *T. brevignatha.* In this case the activity of all individuals of these species observed over a 100-m stretch of forest was monitored hourly throughout the 24-hr period (n = 7 days).

RESULTS

Phylogenetic Analyses

The more conservative 12S mtDNA sequences place the Hawaiian *Tetragnatha* into three clades (Figure 1 [Gillespie et al. 1994]). Because of the use of different data sets to examine the different clades, and the rapid evolution of COI sequences and allozymes, we consider the phylogeny of the two major clades separately: (1) spiny-leg clade, and (2) the complex of web builders. Within each clade, because of the different rates of evolution of the different DNA sequences, the 12S and COI data sets were analyzed separately and the resulting tree topologies compared.

SPINY-LEG CLADE. Figure 3 shows the pattern of phylogeny generated for species within the spiny-leg clade. For the 12S mtDNA sequences we found 2–13 base changes between members of the clade. Figure 3, *A* shows a strict consensus of 55 trees generated based on parsimony (length 55, CI 0.84 including uninformative characters; length 21, CI 0.67 ignoring uninformative characters). For the COI mtDNA sequences we found 28–57 base changes between members of the clade. The single tree generated (length 235, CI 0.81 including uninformative characters; length 194, CI 0.71 ignoring uninformative characters) is shown in Figure 3, *B.* For the allozymes, genetic distances between species were large (Nei's $D > 1$ between *T. pilosa* and both *T. brevignatha* and *T. restricta*). The tree generated is based on genetic distance (Figure 3, *C*). The phylogenetic relationships from the different data sets show considerable agreement, although the extent of congruence could not be measured because the taxa included in the analyses differed considerably. The trees all suggest that within the spiny-leg clade the most ancestral species occur on the oldest island. In contrast to the morphological results (Figure 2), the molecular phylogenies all suggest that the taxa on any

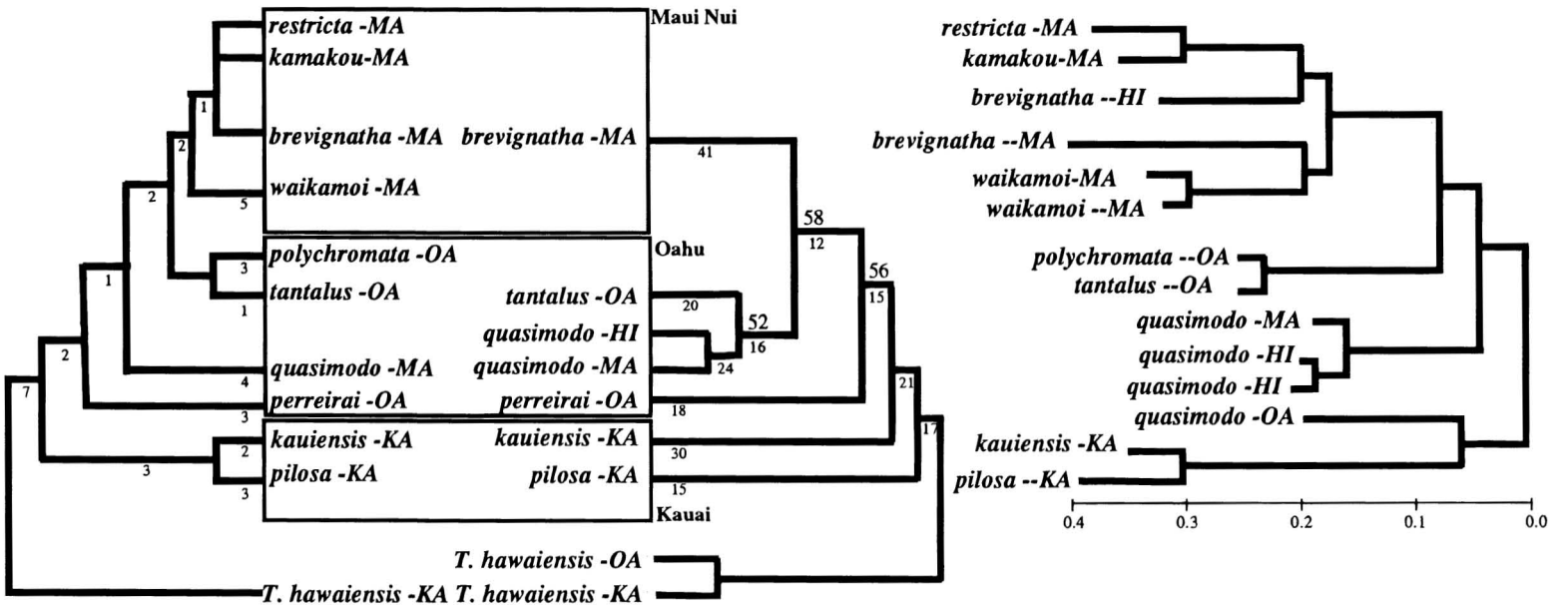

FIGURE 3. Phylogeny of species in the spiny-leg clade. *A*, 12S mtDNA sequences; *B*, COI mtDNA sequences; *C*, Allozymes. For the 12S tree, the strict consensus of 55 trees is shown. For both 12S and COI phylogenies, bootstrap values are given above and adjacent to nodes. Branch lengths are indicated below branches. Boxed areas indicate the islands (Kaua'i, O'ahu, and Maui Nui) on which species occur. Species that occur on more than one island are included in the boxes of the oldest island on which they are found (the oldest point of occurrence frequently marks the point of origination of taxa in the archipelago). For the Distance Wagner tree (*C*), values indicate distance from the root. (See Figure 1 for locality abbreviations.)

one island are generally most closely related to each other.

WEB-BUILDING SPECIES. Figure 4 shows the pattern of phylogeny generated for species within the complex of web-building species. For the 12S mtDNA sequences we found a maximum of 13 base changes between members of the group. The strict consensus of 22 trees is shown in Figure 4, *A* (length 86, CI 0.85 including uninformative characters; length 58, CI 0.64 ignoring uninformative characters). For COI sequences we found up to 80 base changes between members of the group. Even between populations of the same species, we found 9–20 base changes. The single tree generated (length 320, CI 0.84 including uninformative characters; length 280, CI 0.82 ignoring uninformative characters) is shown in Figure 4, *B*. Again, although the patterns generated from the two data sets show general agreement, the extent of congruence could not be measured because of the differences in the taxa included in the analyses.

Characteristics of Representative Species

MICROHABITAT ASSOCIATIONS. Endemic *Tetragnatha* are mostly found in native forest and are largely confined to elevations above 300 m. Except for species in the spiny-leg clade, all species build webs and are often associated with very specific microhabitats (Table 1). Representatives of the *T. hawaiensis* clade are on all islands and, except for Hawai'i (*T. hawaiensis* itself is found at all elevations), are confined to

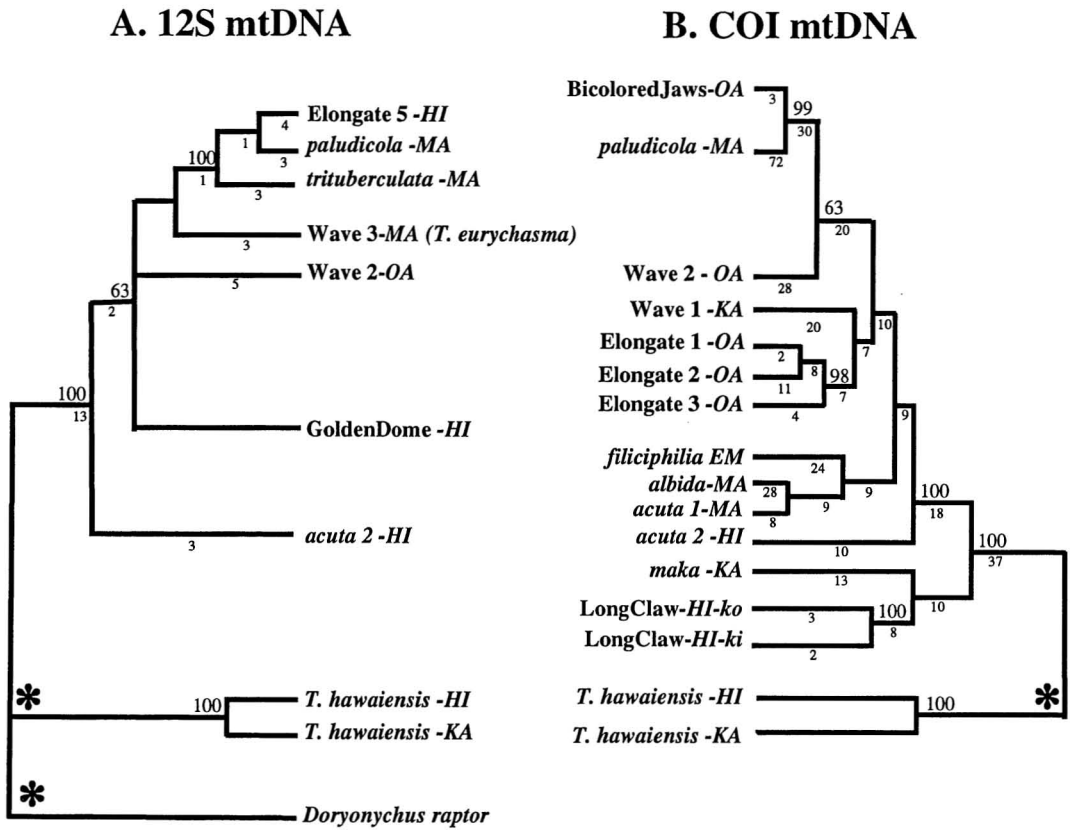

FIGURE 4. Phylogeny of species in the complex of web-building species. *A*, 12S mtDNA sequences; *B*, COI mtDNA sequences. For the 12S tree, the strict consensus of 22 trees is shown. For both 12S and COI phylogenies, bootstrap values are given above and adjacent to nodes. Branch lengths are indicated below branches. * indicates separate colonization events. (See Figure 1 for locality abbreviations.)

TABLE 1

MICROHABITAT SELECTION IN DIFFERENT SPECIES OF HAWAIIAN *Tetragnatha*

GROUP OR COMMON NAME	SPECIES NAME	ISLAND	WEBS BUILT?	HABITAT
	D. raptor	Kaua'i	No web	Below waterfalls, open habitat
	T. hawaiensis	All islands	Webs	Open areas, often near water
Elongate 1		O'ahu	Webs	Open habitat
Elongate 2		O'ahu	Webs	Among leaves
Elongate 3		O'ahu	Webs	In moss
Elongate 4	*T. stelarobusta*	Maui	Webs	Open habitat
Elongate 5		Hawai'i	Webs	Open habitat
Wave 1		Kaua'i	Webs	Open areas, often near water
Wave 2		O'ahu	Webs	Open areas, often near water
Wave 3	*T. eurychasma*	Maui	Webs	Open areas, often near water
	T. acuta 1	Maui	Webs	In bogs (low) or in 'ōhi'a trees
	T. acuta 2	Hawai'i	Webs	On edges of lava flows
	T. albida	Maui	Webs	Open areas in dry forest
	T. maka	Kaua'i	Webs	Cliff edges
	T. filiciphilia	Maui	Webs	Under ferns in forest
	T. paludicola	Maui	Webs	Branches over wet bogs
	T. trituberculata	Maui	Webs	Tree trunks and rocks, wet forest
Golden Dome		Hawai'i	Webs	Open areas in wet forest
Bicolored Jaws		O'ahu	Webs	Low, in moss of bog habitat
Long-clawed		Hawai'i	Webs?	Open areas in wet forest
Spiny-leg 1	*T. kauaiensis*	Kaua'i	Cursorial	Green substrate
Spiny-leg 2	*T. tantalus*	O'ahu	Cursorial	Green substrate
Spiny-leg 3	*T. polychromata*	O'ahu	Cursorial	Green substrate
Spiny-leg 4	*T. waikamoi*	Maui	Cursorial	Green substrate
Spiny-leg 5	*T. brevignatha*	Maui, Hawai'i	Cursorial	Green substrate
Spiny-leg 6	*T. pilosa*	Kaua'i	Cursorial	Brown substrate
Spiny-leg 7	*T. perreirai*	O'ahu	Cursorial	Brown substrate
Spiny-leg 8	*T. quasimodo*	All islands	Cursorial	Brown substrate
Spiny-leg 9	*T. kamakou*	Maui	Cursorial	Brown substrate
Spiny-leg 10	*T. restricta*	Maui, Hawai'i	Cursorial	Brown substrate

elevations below 1000 m. They build orb webs in open areas, up to ca. 4 m above the ground, often in very disturbed forest. *Tetragnatha eurychasma* (Maui) and similar ("Wave") species build small orb webs often with large spaces between the spiral lines and frequently close to water (similar to those of many continental species). *Tetragnatha acuta* and similar species (*T. maka* and *T. albida*) occur in open habitats, often at the edges of cliffs, or forest edges beside lava flows, or in bogs. Species similar to *T. stelarobusta* ("Elongate" species) build orb webs in open areas of native forest, up to ca. 3 m above the ground. *Tetragnatha paludicola* (Maui), *T. trituberculata* (Maui) and "Bicolored Jaws" (O'ahu) are all confined to the wettest sites in mid- to high-elevation native forest.

Representatives of the spiny-leg clade do not build webs. They have been collected from almost all native forest, mostly above 570 m. *Tetragnatha quasimodo,* a robust black/brown spider, occurs on all islands except Kaua'i and is associated with brown substrates (twigs, bark, etc.). *Tetragnatha polychromata, T. tantalus, T. brevignatha,* and *T. waikamoi* are all bright lime-green species and are associated with green substrates (leaves, stems, etc.).

FORAGING BEHAVIOR. Behavioral observation on *T. stelarobusta* revealed that web construction started between 1900 and 2000 hours. The spiders remain on the web through the night, then take in the orb between 0500 and 0600 hours, after which they conceal themselves in the leaf litter. *Tetragnatha eurychasma* often maintains the web through the day and can be found at the hub of the orb in daylight. Species in the spiny-leg clade, which never build webs, act as cursorial predators. There are, however, distinct

differences in the foraging strategy between species in this clade. Comparing *T. quasimodo* and *T. brevignatha,* which coexist on Maui and Hawai'i Island, *T. quasimodo* acts more as an ambush predator, hanging from the vegetation with its front legs held out, whereas *T. brevignatha* adopts a more active hunting strategy, running rapidly over the vegetation (Table 2). Activity of both these species drops in the latter part of the night (midnight to 0600 hours), with inactivity in *T. brevignatha* increasing from 8% to 73%, and in *T. quasimodo* from 10% to 66%. Preliminary observations of prey captured by the different species are indicated in Table 3.

DISCUSSION

The Hawaiian *Tetragnatha* clearly demonstrate considerable deviation from the standard ecology, behavior, and morphology of continental species (Gillespie 1991*a,* 1992*a,* 1993, 1994, Gillespie et al. 1994, Gillespie and Croom 1995). Representatives of the *T. hawaiensis* clade are homogeneous in coloration (all are silver/black) and shape (all are elongate–oval). They are medium-sized spiders that build relatively large webs in open spaces with little evidence of any microhabitat specialization. Prey type depends on the habitat, but because they are often in wet areas, this is generally weak dipterans.

The spiny-leg clade has representatives that show a wide range in color (iridescent lime green to stripes of black and gray), abdomen shape (elongate to diamond-shape), and size (female adult body length ca. 4 mm to ca. 8 mm). They have elongate macrosetae on the first tibia and have never been found to build webs. Their prey are mainly cursorial insects. Based on morphological characters, the "green" species and the "brown" species appear to be most closely related to each other, with differentiation occurring primarily between islands (Figure 2 [Gillespie 1993]). However, although incomplete, each of the molecular data sets examined here (allozymes and 12S and COI mitochondrial DNA) suggests that the species on any one island are generally most closely related to each other (Figure 3). Despite the differences in taxa used for the different data sets, the consistency with which the molecular results yield this pattern suggests that the set of morphological characters uniting the "green" species may have arisen through convergence. At all sites examined to date, there is a single "green" species and usually one "brown" species: Kaua'i (*T. kauaiensis* and *T. pilosa*); O'ahu Wai'anae (*T. polychromata* and *T. quasimodo*); O'ahu Ko'olau (*T. tantalus* and *T. quasimodo*); Maui Nui (*T. waikamoi* and *T. quasimodo,* except for a small area on East Maui where *T. waikamoi* is replaced by *T. brevignatha*); Hawai'i (*T. brevignatha* and *T. quasimodo*). In most populations there is one additional species: on Kaua'i (*T. mohihi*), O'ahu (*T. perreirai*), Maui Nui (*T. kamakou,* except in a small section of East Maui, where *T. restricta* occurs), and Hawai'i (*T. restricta*). All the available molecular information suggests that these sets of three taxa on each island/volcano have, to a large extent, evolved independently. The "green" species are clearly not monophyletic. Neither are *T. kamakou, T. perreirai* and *T. restricta,* or *T. quasimodo* and *T. pilosa.* Only *T. quasimodo* has

TABLE 2

BEHAVIORAL COMPARISON OF TWO DIFFERENT SPINY-LEG SPECIES ON MAUI

SPECIES	HANGING (%)	RUNNING (%)	INACTIVE (%)
Tetragnatha quasimodo[a] (n = 118)	84	8	8
Tetragnatha brevignatha[b] (n = 92)	16	74	10

NOTE: Activity of all individuals observed was recorded over a 5-hr period (1900 to 2400 hours) on 10 different nights between October 1987 and February 1988.

[a] Brown.

[b] Green.

TABLE 3

PREY CAPTURE BY HAWAIIAN *Tetragnatha*

	CURSORIAL ARTHROPODS[a]						FLYING INSECTS[b]			
				LEP. LARVA			LEP. ADULT		TIPULIDAE	
	THER.	THOM.	HOMOPT.			AMPH.		DROS.		
SIZE RANGE (mm)	3–4	2–4	3–5	5–10	10–15	5	5–6	6	2–6	8–10
Spiny-leg clade										
T. brevignatha (n = 10)	0.10	0.10	0.30	0.20	0.20	—	—	—	—	—
T. waikamoi (n = 8)	—	—	0.50	0.25	0.25	—	—	—	—	0.10
T. kauaiensis (n = 8)	0.25	—	0.63	—	—	—	—	—	0.12	0.10
T. kamakou (n = 4)	0.60	—	—	—	0.40	—	—	—	—	—
T. quasimodo (n = 11)	0.09	0.09	0.09	0.27	0.36	—	—	0.09	—	—
Web-building species										
T. stelarobusta (n = 10)	—	—	—	—	—	—	0.90	—	—	0.10
T. hawaiensis (n = 8)	—	—	—	—	—	—	0.12	—	0.12	0.76
T. paludicola (n = 6)	—	—	—	—	—	—	—	—	0.17	0.83
T. trituberculata (n = 4)	—	—	—	—	—	—	—	0.75	—	0.25
"Bicolored Jaws" (n = 2)	—	—	—	—	—	1.00	—	—	—	—
T. eurychasma (n = 12)	—	—	—	—	—	—	—	—	0.33	0.67

[a] Spiders in the families Theridiidae (Ther.) and Thomisidae (Thom.), insects in the order Homoptera (Homopt.), larval lepidopterans (Lep. larva), and crustacean amphipods (amph.).

[b] Adult lepidopterans (Lep. adult) and dipterans in the families Drosophilidae (Dros.) and Tipulidae.

established itself widely through the archipelago; the other species appear to have differentiated independently into ecological counterparts on each island or volcano.

An alternative explanation for the discrepancy between the morphological and molecular (mtDNA) phylogenies might be that hybridization among species within islands has occurred, resulting in a similar mtDNA haplotype among these species. However, the relationships indicated by the allozymes (Figure 3, *C*) shows a pattern similar to that for mtDNA, so argues against this explanation.

Representatives of the large complex of web-building species exhibit an enormous range in color (shiny bottle green to white with patterns of many different colors and forms), body shape (elongate to almost round, smoothly contoured, angular, or tuberculate), and size (female adult body length ca. 2 mm to ca. 8 mm). Webs are built in microhabitats that are highly species specific, and coloration of the spiders appears to be finely tuned to their selection of microhabitat. *Tetragnatha stelarobusta* captures primarily moths, as might be expected from its selection of a nocturnal web site in relatively dry, open sites in the forest. Those species that select sites close to water or in root crevices, such as *T. eurychasma* (and other "Wave" species) and *T. paludicola,* prey primarily on tipulids. *Tetragnatha trituberculata,* with its web close to very wet tree bark, captures mostly drosophilids. Although only two captures were observed for "Bicolored Jaws" from O'ahu, this species appears to be specialized for feeding on terrestrial amphipods: It has lost the cheliceral teeth characteristic of *Tetragnatha,* instead having minute serrations along the margins of the chelicerae. It is likely that the serrations may serve to hold the amphipod prey in a manner similar to that of the beaks of many piscivorous birds.

Molecular information, although clearly preliminary, indicates that many of the apparent morphological "clades" in the large group of web-building species of Hawaiian *Tetragnatha* are paraphyletic (Figure 4). In particular, species similar to *T. acuta* are found on several different islands: *T. maka* on Kaua'i, *T. acuta* and *T. albida* on Maui, and *T. acuta* on Hawai'i. The molecular evidence suggests that these species form a paraphyletic group. Likewise, species similar to *T. eurychasma* appear also to be paraphyletic. It

may be that, as these species have colonized the islands, they have differentiated to form new species on the different islands, but the ancestral species remains, and in similar form, on each island.

The extent to which communities in similar environments converge in structure has been debated extensively for many years (Orians and Paine 1983, Schluter 1986, Ricklefs and Schluter 1993). Various mechanisms have been proposed to account for consistent variation that is often observed among species in different communities. However, studies disagree on the extent to which observed differences among species in a given community evolved in allopatry or sympatry. For the finches of the Galápagos, Grant (1986) proposed that differences among species evolve in allopatry and subsequent character displacement causes rapid divergence in feeding structures between the species when they come together in sympatry.

The results of our study suggest that differences among species of Hawaiian *Tetragnatha* evolved largely on the same island. A similar phenomenon has been found among species of *Anolis* in the Caribbean (Losos 1992, Losos et al. 1994). *Anolis* species on the different islands have arisen as a result of one-to-one convergence of the same set of "ecomorph" types on each island (Losos 1992). In addition, the same set of "ecomorphs" appear repeatedly on the different islands and have even arisen through similar evolutionary sequences. In a similar manner, at least the "green" and "brown" species of spiny-leg Hawaiian *Tetragnatha* seem to be ecomorphs that have arisen independently on the different islands.

Another trend in the Hawaiian *Tetragnatha* that is reminiscent of the Caribbean *Anolis* is that evolution may show a similar progression toward specialization: For the *Anolis,* no instances have been found in which taxa have reverted to a more generalized condition (Losos et al. 1994). The trend toward specialization is reminiscent of Wilson's "taxon cycle" (Wilson 1961) and Erwin's "taxon pulse" (Erwin 1979, 1985) in which there is a largely irreversible shift toward habitat specialization during speciation episodes (Wilson 1961, Erwin 1985). For Melanesian ants Wilson (1961) proposed that widespread dispersive populations give rise to more restricted and specialized species, and subsequent divergence leads to local endemics. During this process a lineage proliferates from a single generalist species to many specialized species. Although the results presented here for the web-building species of Hawaiian *Tetragnatha* are preliminary, they do suggest that the more generalist species (in terms of foraging behavior) are more ancestral to species with very specialized foraging behaviors and microhabitat associations.

In conclusion, our study provides a preliminary assessment of phylogeny for two clades of Hawaiian *Tetragnatha.* The results, if substantiated by future analyses, suggest that the adaptive radiation of the Hawaiian *Tetragnatha* has occurred via sequential microhabitat partitioning following colonization of an island in both web-building and nonweb-building clades. Niche partitioning appears to have taken place repeatedly among species groups on each island, resulting in independently evolved ecological equivalents. Microhabitat specialization is most evident in the web-building species and may occur progressively with speciation on any one island.

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LITERATURE CITED

BRISTOWE, W. S. 1958. The world of spiders. Collins, London.

CARSON, H. L., and D. A. CLAGUE. 1995. Geology and biogeography of the Hawaiian Islands. Pages 14–29 *in* W. L. Wagner and V. A. Funk, eds. Hawaiian biogeography: Evolution on a hot spot archipelago. Smithsonian Institution Press, Washington.

CLARY, D. O., and D. R. WOLSTENHOLME. 1985. The mitochondrial DNA molecule of *Drosophila yakuba*: Nucleotide sequence, gene organization and genetic code. J. Mol. Evol. 22:252–271.

COOKE, C., J. MONTAGUE, and Y. KONDO. 1960. Revision of Tornatellinidae and Achatinellidae (Gastropoda, Pulmonata). Bernice P. Bishop Mus. Bull. 221:1–303.

CROOM, H. B., R. G. GILLESPIE, and S. R. PALUMBI. 1991. Mitochondrial DNA sequences coding for a portion of the RNA of the small ribosomal subunits of *Tetragnatha mandibulata* and *Tetragnatha hawaiensis* (Araneae, Tetragnathidae). J. Arachnol. 19:210–214.

DESALLE, R. 1995. Molecular approaches to biogeographic analysis of Hawaiian Drosophilidae. Pages 72–89 *in* W. L. Wagner and V. A. Funk, eds. Hawaiian biogeography: Evolution on a hot spot archipelago. Smithsonian Institution Press, Washington.

ERWIN, T. L. 1979. Thoughts on the evolutionary history of ground beetles: Hypotheses generated from comparative faunal analyses of lowland forest sites in temperate and tropical regions. Pages 539–592 *in* T. L. Erwin, G. E. Ball, D. R. Whitehead, and A. L. Halpern, eds. Carabid beetles: Their evolution, natural history and classification. W. Junk, The Hague.

———. 1985. The taxon pulse: A general pattern of lineage radiation and extinction among carabid beetles. Pages 437–472 in G. E. Ball, ed. Taxonomy, phylogeny and zoogeography of beetles and ants. W. Junk, Dordrecht.

FREED, L. A., S. CONANT, and R. C. FLEISCHER. 1987. Evolutionary ecology and radiation of Hawaiian passerine birds. Trends Evol. Biol. 2(7): 196–203.

GILLESPIE, R. G. 1987. The mechanism of habitat selection in the long jawed orb weaving spider *Tetragnatha elongata* (Araneae, Tetragnathidae). J. Arachnol. 15:81–90.

———. 1991*a*. Hawaiian spiders of the genus *Tetragnatha:* I. Spiny leg clade. J. Arachnol. 19:174–209.

———. 1991*b*. Predation through impalement of prey: The foraging behavior of *Doryonychus raptor* (Araneae, Tetragnathidae). Psyche (Camb.) 98:337–350.

———. 1992*a*. Hawaiian spiders of the genus *Tetragnatha* II. Species from natural areas of windward East Maui. J. Arachnol. 20:1–17.

———. 1992*b*. Impaled prey. Nature (Lond.) 355:212–213.

———. 1993. Biogeographic pattern of phylogeny among a clade of endemic Hawaiian spiders (Araneae, Tetragnathidae). Mem. Queensl. Museum 33:519–526.

———. 1994. Hawaiian spiders of the genus *Tetragnatha:* III. *T. acuta* clade. J. Arachnol. 22:161–168.

GILLESPIE, R. G., and H. B. CROOM. 1995. Comparison of speciation mechanisms in web-building and non-web-building groups within a lineage of spiders. Pages 121–146 *in* W. L. Wagner and V. A. Funk, eds. Hawaiian biogeography: Evolution on a hot spot archipelago. Smithsonian Institution Press, Washington.

GILLESPIE, R. G., H. B. CROOM, and S. R. PALUMBI. 1994. Multiple origins of a spider radiation in Hawaii. Proc. Natl. Acad. Sci. U.S.A. 91:2290–2294.

GRANT, P. R. 1986. Ecology and evolution of Darwin's finches. Princeton University Press, Princeton, New Jersey.

HEBERT, P. D. N., and M. J. BEATON. 1989. Methodologies for allozyme analysis using cellulose acetate electrophoresis. Helena Laboratories, Beaumont, Texas.

HORMIGA, G., W. G. EBERHARD, and J. A. CODDINGTON. 1995. Web construction behavior in Australian *Phonognatha* and the phylogeny of nephiline and tetragnathid spiders (Araneae, Tetragnathidae). Aust. J. Zool. 43:313–343.

KANESHIRO, K. Y., and L. V. GIDDINGS. 1987. The significance of asymmetrical sexual isolation and the formation of new species. Evol. Biol. 21:29–43.

KANESHIRO, K. Y., R. G. GILLESPIE, and H. L. CARSON. 1995. Chromosomes and male genitalia of Hawaiian *Drosophila:* Tools for

interpreting phylogeny and geography. Pages 57–71 *in* W. L. Wagner and V. A. Funk, eds. Hawaiian biogeography: Evolution on a hot spot archipelago. Smithsonian Institution Press, Washington.

KARSCH, F. 1880. Sitzungs-Berichte der Gesellschaft Naturforschender freunde zu Berlin. Jahrgang. Sitzung vom 18:76–84.

KASTON, B. J. 1948. How to know the spiders, 3rd ed. Wm. C. Brown Co., Dubuque, Iowa.

LEVI, H. W. 1981. The American orb-weaver genus *Dolichognatha* and *Tetragnatha* north of Mexico (Araneae: Araneidae, Tetragnathinae). Bull. Mus. Comp. Zool. Harv. Univ. 149(5): 271–318.

LOSOS, J. B. 1992. The evolution of convergent structure in Caribbean *Anolis* communities. Syst. Biol. 41:403–420.

LOSOS, J. B., D. J. IRSCCHICK, and T. W. SCHOENER. 1994. Adaptation and constraint in the evolution of specialization of Bahamian *Anolis* lizards. Evolution 48:1786–1797.

MAYR, E. 1963. Animal species and evolution. Harvard University Press, Cambridge, Massachusetts.

OKUMA, C. 1988. Redescriptions of the Hawaiian spiders of *Tetragnatha* described by Simon (Araneae, Tetragnathidae). J. Fac. Agric. Kyushu Univ. 33:77–86.

———. 1990. *Doryonychus raptor* (Araneae, Tetragnathidae), an interesting Hawaiian spider. Esakia, special issue 1:201–202.

ORIANS, G. H., and R. T. PAINE. 1983. Convergent evolution at the community level. Pages 431–458 *in* D. J. Futuyma and M. Slatkin, eds. Coevolution. Sinauer, Sunderland, Massachusetts.

OTTE, D. 1994. The crickets of Hawaii: Origin, systematics and evolution. The Orthopterists' Society: Academy of Natural Sciences of Philadelphia, Pennsylvania.

PERKINS, R. C. L. 1913. Introduction. *In* D. Sharp, ed. Fauna Hawaiiensis, Vol. 1:15–228. Cambridge University Press, Cambridge.

PLATNICK, N. I. 1993. Advances in spider taxonomy 1988–1991. New York Entomological Society and American Museum of Natural History, New York.

RICHARDSON, B. J., P. R. BAVERSTOCK, and M. ADAMS. 1986. Allozyme electrophoresis. A handbook for animal systematics and population studies. Academic Press, New York.

RICKLEFS, R. E., and D. SCHLUTER. 1993. Species diversity in ecological communities: Historical and geographical perspectives. University of Chicago Press, Chicago.

SCHLUTER, D. 1986. Tests for similarity and convergence of finch communities. Ecology 67:1073–1085.

SHAW, K. L. 1995. Biogeographic patterns of two independent Hawaiian cricket radiations (*Laupala* and *Prognathogryllus*). Pages 39–56 *in* W. L. Wagner and V. A. Funk, eds. Hawaiian biogeography: Evolution on a hot spot archipelago. Smithsonian Institution Press, Washington.

———. 1996. Sequential radiations and patterns of speciation in the Hawaiian cricket genus *Laupala* inferred from DNA sequences. Evolution 50:237–255.

SIMON, C. 1987. Hawaiian evolutionary biology: An introduction. Trends Ecol. Evol. 2:175–178.

SIMON, C., F. FRATI, A. BECKENBACH, B. CRESPI, H. LIU, and P. FLOOK. 1994. Evolution, weighting, and phylogentic utility of mitochondrial gene sequences and a compilation of conserved polymerase chain reaction primers. Ann. Entomol. Soc. Am. 87: 651–701.

SIMON, E. 1900. Arachnida. Pages 443–519, *in* Fauna Hawaiiensis 2(5), pls. 15–19

SWOFFORD, D. L. 1993. PAUP. Phylogenetic analysis using parsimony, version 3.1.1. Smithsonian Institution, Washington.

TAMURA, K. 1992. Estimation of the number of nucleotide substitutions when there are strong transition-transversion and G+C-content biases. Mol. Biol. Evol. 9:678–687.

TARR, C. L., and R. C. FLEISCHER. 1995. Evolutionary relationships of the Hawaiian honeycreepers (Aves, Drepanidinae). Pages 147–159 *in* W. L. Wagner and V. A. Funk, eds. Hawaiian biogeography: Evolution on a hot spot archipelago. Smithsonian Institution Press, Washington.

WILSON, E. O. 1961. The nature of the taxon cycle in the Melanesian ant fauna. Am. Nat. 95:169–193.

APPENDIX 1

ALIGNED SEQUENCES FOR RIBOSOMAL 12S MITOCHONDRIAL DNA

(Matched characters indicated by .; gaps indicated by *; missing characters indicated by X. See Figure 1 for locality abbreviations.)

```
Elongate 5 HI      CAC*TTTTAATTATATTTTATATACCGCCGTCTTGAATAGATCATAAA*ATTTTGTTCAAAATAAAATTCTAAAAATTTAGGTAAAGGTGTAAACTATCTAAAA
Golden Dome HI     ...*.....................................*.....................................................
acuta 2 HI         ...*.....................................*.....................................................
paludicola MA      ...*.....................................T.....*.....................................................T.C...
eurychasma MA      ...*.....................................C.........*.........G.....................................................T.....
trituberculata MA  ...*.....T.....................G.......*.....................................................G.
Wave 2 OA          XX.*.........C.....................................*.....................................................TC....

restricta MA       ...*.....................................C.TA...*.....A.......A.....................................T.....
brevignatha MA     ...*.....T.....................................C.TA...*.....A.......A.....................................G.....
waikamoi MA        ...*.....C.....................................C.TA...*.....A.......G.....................................T.....
kamakou MA         ...*......C.....................................C.TA...A.....A.......G.....................................T.....
quasimodo MA       ...*.C.......C.....................................TA...*.....A.......A.........G.....................T.....
perreirai OA       ...*.....................................TTA...*.....A.......A.....................................T.....
tantalus OA        ...T.....................................C.TA...*.....A...T...A.....................................XXXXXXXXXX
polychromata OA    ...T.........C.....................................C.TA...*.....A.......A.....................................T.....
kauaiensis KA      ...*.....................................T....A.....A.....................C.....................T.....
pilosa KA          ...*.....................................CA...*.....A.....................................T.....

hawaiensis HI      ...*.......A.....................T.A.....A..A.A...*.....A...........T.....G.....................A...G.
hawaiensis KA      ...*.......A.....................A.....A..A.A...*.....A...........T.....G.....................A...G.
Doryonychus
   raptor KA       T..*..GG.T.............T.T....A.C.....T...TA..T*..A..A....C.....T...A.....................G....CA.CC..


Elongate 5 HI      GATAAAATGTATTACACTAAAAAAAATTTAAGGATCAAATTAAAATATAAT TTAAGAAAGGGGATTTATAAG*TATTATTTAATCAAAATTAAAATTTGCCA
Golden Dome HI     ................T.....................................T.........A...........*.........T.....................
acuta 2 HI         ...G............T.....................................T.........AA..........*...........T.....................
paludicola MA      ...G............T.....................................*.....................*.....................GC
eurychasma MA      ...G............T.....................................TC........A...........*.....................GC
trituberculata MA  ...G............T.....................................*.........A...........*.....................GC
Wave 2 OA          ................T....T.....................G.........TC.......AA..........*...........T..............GG

restricta MA       ................T....T.............G.....*..C....CC.C......AA..........*...........T..T...........A.C
brevignatha MA     ................T....T.............G.....*..C....CT.C......AA..........*...........T..T...........A.C
waikamoi MA        ................T....T.............GG....*.........T.C......AA..........*...........T..T...........A.C
kamakou MA         ................T....T.............G.....*..C....CC.CG.....AA..........*...........T..T...........A.C
quasimodo MA       ................T....T.............G.....*.........C.C......AA..........*...........T..T...........A.G
perreirai OA       ..C.............T....T.............G.G...*.........T.C......AA..........*...........T..T...........A.G
tantalus OA        XXXXXXXXX
```

APPENDIX 2

ALIGNED SEQUENCES FOR CYTOCHROME OXIDASE MITOCHONDRIAL DNA

(Matched characters indicated by .; missing characters indicated by -. See Figure 1 for locality abbreviations.)

Appendix 2A

```
pilosa KA       ------------------------------------------------------------------------------------------------------------
quasimodo MA    ----------------------TCGAATAAATAATTTAAGATTTTGGCTTCTCCCTCCTTCCCTTTTTATGTTATTTATTTCATCTATAGTAGATGTGGGAGTTGGA
kauiensis KA    --------CGAT-T-G-GGG-CCGGGGATAAATAATTTAAGATTTTGACTTCT-CCCCCCTCTCTTTTTTATATTATTTATCTCATCTATAGTAAATGTTGGAGTGGGG
tantalus OA     ------------------------------------------------------CT-CCCCCCTCTCTTTTTTATATTATTCATTTCATCTATAGTAAATGTGGGAGTTGGG
brevignatha MA  --------------------CCTCGAATAAATAATATAAGATTTTGAATTCTTCCCCCTTCCCTTTTTAT-TTATTTATCTCATCTATAG-GGAT-TGGGAGTGGG-
perreirai OA    ------------------------------------------------------------------------------------------------------------
qusimodo HI     ----------------------GCGAATAAATAATTTGAGATTTTGACTTCTTCCTCCTTCTCTTTTTATATTATTTATTTCGCCTATAGTAGACGTAGGGGTTGGG
hawaiensis HI   ----------------------------TAAATAATTTAAAATTTTGG-GA-GC-CCCGTCCCTTGT-TATATTACTTATTTCA-CTATAGTGGATGTTGGGGTTGGA
hawaiensis KA   ------------------------------------------------------GCTCTTACT-TATTACT-ATA-CAT-TATAGTGGAT-TTGGAGTTGGA

pilosa KA       ------------------------------------GGCATCCAGGGAGATCTGTAGATTTTGCAATTTTTTCGCTTCATTTAGCGGGGGCTTCATCTATTATA
quasimodo MA    TCAGGGTGGACTGTGTATCCTCCTTTGGCA-CCCTGGATG....C.....................................A.........A........C.........
kauiensis KA    GCGGGATGAACTGTGTACCCCCCCTTA-CGTCCCTGGACG.A.....G.............A..............-.A.-........A.....A..T.......-G
tantalus OA     GCAGGTTGGACTGTATATCCCCCTTTAGCATCCCTGGATG.....................G.................A...........A........C-........
brevignatha MA  GCGG-ATGGACTG-ATACCCCCC-TTA-GATCTT-AGATA........A.T.......AG.-.............A.....C.............T.........
perreirai OA    ------------------------------------GATG.-.....G..........G.................A......C....A........T.........
qusimodo HI     TCAGGGTGGACTGTGTACCCTCCTTTAGCATCTTTAGATG.C.....................G.....G.............A.........A.......C.........
hawaiensis HI   -CAGGGTGAACTGTATACCCACCCCTGGCGGCCCTTAAGG.C......-...A.......A.......T.....-..T.....-.....A..T.....T-........
hawaiensis KA   GCAGGGTGAACTGT-TATCCACCCCTG-C-GC-CTTGTGG.T.....................A.......G..T.....CC.T..A...-...-A....T...-.-.......

pilosa KA       GGTGCTATTAATTTTATTTCTACAATTATTAATATGCGTATGAGAGGGGTTTCTATGGAAAAGGTCCCCCTT-T-G-G-GATC-GTTTTAATTAC-GCAGTAT-GCTT
quasimodo MA    ..A.....................................C..A..A..A.........A....A..G..A.........T.T.T.T.G..T...........G.......TAT.A
kauiensis KA    ..G..............-C.....................-.....A.A...AA.C.....A-.G.....-..A...T.T.T.T....T...........T.....T.GA...
tantalus OA     ..G..A.....................................A..G..A.-.......A.C..A-....A..T.....-.T.-.T.T....T...........A.....T.TA..A
brevignatha MA  .....................C..............A..A..A........-.G.G..A..G........T.-CT.A.TAA....A...........A.......TAT.A
perreirai OA    ..G..A--.............C...........C..A.....A.......G-.....A..G..A..G..T.CCC.T.T.T....T.....G.....A.......TA...
qusimodo HI     ..G........C.....................-...A..A..A.........A....A.....A..T......T.C.T.T.G..T...........G.-...-CTA..A
hawaiensis HI   ..G...........C..-.....T...........A..A-.A.A...T...-.........G......T..-T.G.T.T....T..--.........-.....G.T.T.A
hawaiensis KA   ..G...........C...A.-..T...........A..A-.A......A.....-....-.....T..AT.G.T.T....T..AGG......A.-...-.T-..A
```

Appendix 2B

```
maka KA         ----------------GGGACCGCGAATAAATAACTTAAGATTTTGGT-ACTTCCTCCTTCTCTTTTTATATTATTTATTTCATCTATAGTGGATGTGGGGGTTGGA
acuta MA        ------------------------------....T.....-..C...T.....C.....A.....................T.........A.....A........G
albida MA       ------------------------CGCG..T..T..TTT...A.T-GGGAC.T.C.....A.....................T-.....A.AA..-.AT.......G
acuta HI        ----------------------...T.-.....T.............-----C..C.....C.....................T.........AA....A........G
LongClawed HI ki ---------------------...G....T--.--.....-.........A.....C.....A...........G.........-.C...-.............A...
LongClawed HI ko ----------------.......G.....................T.....C.....A...........G.........C.....................A...
Elongate1 OA    ----------------.......G.T..---...T.........-.A.T.....C.....A...........G.........C.........AA....A........G
Elongate2 OA    ------------------------------------------------------------------------G..-.....C.AA.....A..G.-A........G
Elongate3 OA    ----------------.......G.T........T....-........T.....C...-.A.....-....G.........C.........AA....A........G
Wave1 KA        ------------------------------------------------------------....G-.............T.....G..AA....T.....G
Wave2 OA        ------CCAGATATGGCTTTT.....T........T.........-...TG.........A.A...A.......G........T.......-A.....A........G
filiciphilia MA ------------------------------------------------C.....A...G....G.C...C..C..T.....A.....A........G
BicoloredJaws OA ------------------------------------.....-..-...GG.....C.--..AG.CCC....C.G.....T..A.....A.....A........G
paludicola MA   -------TGAATAAATCCC.-GTA.C..A..TCCCAAG.TTTG-G.TTGGT.CC..C-T--GGT.C..C.CCC.G.AG.-GG--C.A.A...A.....A.-.C.--.G

maka KA         GCAGGATGAACTGTTTATCCTCCTTTAGCATCCTTGGATGGACACCCAGGGAGATCTGTGGATTTTGCAATTTTTTCTCTTCATTTAGCAGGGCCTCTTCTATTATA
acuta MA        .....G.........C..C......C.T.....T.......G..T.....T.....................................................T.....C......
albida MA       ...A.G........C..C......C.T.....T...........T...A.T.....................................-..............T.....C......
acuta HI        .....G.................C.T.....T..A.....G..T.....T.....................T.A.....G.........T.............
LongClawed HI ki -....G.............C...C.T.....T.........G..T..G..C.A...C.....................T.A.....-....A..T.............
LongClawed HI ko .....G.............C...C.T.....................................................T.A.....-....A..T.............
Elongate1 OA    .....G.............C...C.T.....T.........G..T..G..C.A...C.....................-.....-.....T.....A......
Elongate2 OA    .....G....-.........C.T.C.T.....T.........G..T..G..C.....C.....................-.....-.....T.T...A......
Elongate3 OA    .....G.............-...C.T.....T.........G..T..G..T.A.....................................-.....TC....A......
Wave1 KA        .....G.........-..C...C...C.T..G..T..A-.C..G..T..G.....................G.....-.....-.....G.....T..C.........
Wave2 OA        .............C.....C...C.T.G...T.........-.......T.....-....-.........GA-....C.....-.....-T..C.........
filiciphilia MA .....................C..CC.T..C..T..A..G..G..T..G..T.....................C.....-.....T.....-.....G
BicoloredJaws OA .............C.....C...C.T..G..T.....................T.....-.....-.....-.....-.....-.....T..C.........
paludicola MA   C....T...C....CA....C...CATC.G...-GT.G......G-.-..T.............A.-.....-.....-.A.....C...-....-T-.C.....-.

maka KA         GGAGCTATTAATTTTATTTCTACCATTTTAAATATACGGATAAGAGGGGTTTCTATAGAAAAGGTACCTTTATTTGTGTGATCAGTTTTAATTACTGCTGTATTACTT
acuta MA        ..G.....................T.....T.........A.....................G..C..T..G.....T..-.....................T.A
albida MA       ..G..C....-.............T..-.-T.......CA...-.....-.....-...-..G...C-T.-.....-.....A.....-.-..G...T-A
acuta HI        ..G.....................T.....T.........A.....................C..CC.T.....-.....T.....-.....C..T.A
LongClawed HI ki ..G.....................T.....-.....-.....-.....-.....-.....T.....-.....-.....C..T.A
LongClawed HI ko ..G.....................T.....-.....-.....-..-.....-.....-.T.....-.-.....-GGA
Elongate1 OA    ..G.....................T.....T....-.-.....-.....G.....C..CC.T.....-.....T.....C.....C.....T.A
Elongate2 OA    ..G..T.----------.....T.....T.....G.....-.....G..--C..T..CC.T.....-.....C.-...C.....T.A
Elongate3 OA    ..G.....................T....-.T....-.....-.....-.....C..CC.T.....A.....T.....C.....G.-.TAA
Wave1 KA        ..T.....................T...G.T.....-.....C..A.....-.....A..C..AC.T.....A.....T.....C.....A..G...T.A
Wave2 OA        ..G.........G--.......T..-GG..-......A.....-.....-.....-C..-C.T-...C.....T-C-.C-.....-.G--..-GA
filiciphilia MA ..G.....................T.....A.....-.....TA.....G..CC.T.....-.....T..G.A..A.....-.....G..T.A
BicoloredJaws OA ..G.....................T.....T.....A.....-.....-.....T...C.T.....-.....T.....-.....GC..T.A
paludicola MA   ..GC.-.C.....-...C.T.....T..A.GC..-.....-.....-.....A..-CGG..-T...CAT.....-.....T.-G.....C..C....GC-.T.A
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